PATENT

DOCKET NO.: UBCV-0006/01-010
Application No.: 10/039,760

This listing of claims will replace all prior versions, and listings, of claims in the application.

Listing of Claims:

1-32. Cancelled.

33. (Previously Presented) A method for reducing colonization of enterohemorrhagic *Escherichia coli* (EHEC) in a non-human mammal comprising administering to said non-human mammal an effective amount of a composition comprising an isolated EHEC cell culture supernatant, wherein the cell culture supernatant is produced by the process of incubating the cell culture in media comprising minimal media supplemented with 0.1% Casamino Acids, 0.4% glucose, 8 mM $MgSO_4$ and 44 mM $NaHCO_3$.

34. (Previously Presented) A method for reducing shedding of enterohemorrhagic *Escherichia coli* (EHEC) from a non-human mammal comprising administering to said non-human mammal an effective amount of a composition comprising an isolated EHEC cell culture supernatant, wherein the cell culture supernatant is produced by the process of incubating the cell culture in media comprising minimal media supplemented with 0.1% Casamino Acids, 0.4% glucose, 8 mM $MgSO_4$ and 44 mM $NaHCO_3$.

35. (Previously Presented) The method of claim 33, wherein the non-human mammal is a ruminant.

36. (Previously Presented) The method of claim 35, wherein the ruminant is a bovine subject.

37. (Previously Presented) The method of claim 33, wherein the composition further comprises an immunological adjuvant.

38. (Previously Presented) The method of claim 33, wherein the EHEC is EHEC O157:H7.

39. (Previously Presented) The method of claim 33, wherein the EHEC is EHEC O157:NM.

40. (Previously Presented) The method of claim 37, wherein the immunological adjuvant comprises an oil-in-water emulsion.

41. (Previously Presented) The method of claim 37, wherein the immunological adjuvant comprises a mineral oil and dimethyldioctadecylammonium bromide.

42. (Previously Presented) The method of claim 37, wherein the immunological adjuvant comprises a non-oil-in-water emulsion.

43. (Previously Presented) The method of claim 37, wherein the immunological adjuvant is present in the composition at a concentration of 20% to 40% (v/v).

44. (Previously Presented) The method of claim 43, wherein the immunological adjuvant is present in the composition at a concentration of 30% (v/v).

45. (Previously Presented) The method of claim 33, wherein the composition further comprises one or more recombinant or purified EHEC antigens selected from the group consisting of EspA, EspB, EspD, Tir and Intimin.

46. (Previously Presented) The method of claim 45, wherein EspA+Tir comprise 10% to 50% of EHEC protein in the cell culture supernatant present in the composition.

47. (Previously Presented) The method of claim 37, wherein the composition further comprises one or more recombinant or purified EHEC antigens selected from the group consisting of EspA, EspB, EspD, Tir and Intimin.

48. (Previously Presented) The method of claim 47, wherein EspA+Tir comprise 10% to 50% of EHEC protein in the cell culture supernatant present in the composition.

49. (Previously Presented) The method of claim 34, wherein the non-human mammal is a ruminant.

50. (Previously Presented) The method of claim 49, wherein the ruminant is a bovine subject.

51. (Previously Presented) The method of claim 34, wherein the composition further comprises an immunological adjuvant.

52. (Previously Presented) The method of claim 34, wherein the EHEC is EHEC O157:H7.

53. (Previously Presented) The method of claim 34, wherein the EHEC is EHEC O157:NM.

54. (Previously Presented) The method of claim 51 , wherein the immunological adjuvant comprises an oil-in-water emulsion.

55. (Previously Presented) The method of claim 51, wherein the immunological adjuvant comprises a mineral oil and dimethyldioctadecylammonium bromide.

56. (Previously Presented) The method of claim 51 , wherein the immunological adjuvant comprises a non-oil-in-water emulsion.

57. (Previously Presented) The method of claim 51, wherein the immunological adjuvant is present in the composition at a concentration of 20% to 40% (v/v).

58. (Previously Presented) The method of claim 51, wherein the immunological adjuvant is present in the composition at a concentration of 30% (v/v).

59. (Previously Presented) The method of claim 34, wherein the composition further comprises one or more recombinant or purified EHEC antigens selected from the group consisting of EspA, EspB, EspD, Tir and Intimin.

60. (Previously Presented) The method of claim 59, wherein EspA+Tir comprise 10% to 50% of EHEC protein in the cell culture supernatant present in the composition.

61. (Previously Presented) The method of claim 51, wherein the composition further comprises one or more recombinant or purified EHEC antigens selected from the group consisting of EspA, EspB, EspD, Tir and Intimin.

62. (Previously Presented) The method of claim 61, wherein EspA+Tir comprise 10% to 50% of EHEC protein in the cell culture supernatant present in the composition.

63. (Previously Presented) The method of claim 37, wherein the immunological adjuvant comprises an agent selected from the group consisting of an emulsifying agent, a muramyl dipeptide, an aqueous agent, a chitosan-based agent, a saponin, an oil, a lipopolysaccharide, a bacterial cell wall extract, a bacterial DNA, a bacterial complex, a synthetic oligonucleotide, and a aliphatic nitrogenous base.

64. (Previously Presented) The method of claim 63, wherein the emulsifying agent is selected from the group consisting of a natural emulsifying agent, a synthetic emulsifying agent, an anionic emulsifying agent, a cationic emulsifying agent, and a nonionic agent.

65. (Previously Presented) The method of claim 64, wherein the natural emulsifying agent is selected from the group consisting of acacia, gelatin, lecithin, and cholesterol.

66. (Previously Presented) The method of claim 64, wherein the anionic emulsifying agent is selected from the group consisting of a potassium salt of lauric acid, a potassium salt of oleic acid, a sodium salt of lauric acid, a sodium salt of oleic acid, an ammonium salt of

lauric acid, an ammonium salt of oleic acid, a calcium salt of a fatty acid, a magnesium salt of a fatty acid, an aluminum salt of a fatty acid, a metallic soap, and an organic sulfonate.

67. (Previously Presented) The method of claim 66, wherein the organic sulfonate is sodium lauryl sulfate.

68. (Previously Presented) The method of claim 64, wherein the cationic emulsifying agent is ceryltrimethylammonium bromide.

69. (Previously Presented) The method of claim 64, wherein the synthetic agent is selected from the group consisting of a glyceryl ester, a polyoxyethylene glycol ester, a polyoxyethylene glycol ether, and a sorbitan fatty acid ester.

70. (Previously Presented) The method of claim 69, wherein the glyceryl ester is glyceryl monostearate.

71. (Previously Presented) The method of claim 69, wherein the sorbitan fatty acid ester is selected from the group consisting of a sorbitan monopalmitate and polyoxyethylene derivatives thereof.

72. (Previously Presented) The method of claim 69, wherein the polyoxyethylene derivatives is polyoxyethylene sorbitan monopalmitate.

73. (Previously Presented) The method of claim 63, wherein the aqueous agent is aluminum hydroxide.

74. (Previously Presented) The method of claim 63, wherein the oil is selected from the group consisting of a mineral oil, a vegetable oil, and an animal oil.

75. (Previously Presented) The method of claim 74, wherein the vegetable oil is selected from the group consisting of canola oil, almond oil, cottonseed oil, corn oil, olive oil, peanut oil, safflower oil, sesame oil, and soybean oil.

76. (Previously Presented) The method of claim 74, wherein the animal oil is selected from the group consisting of cod liver oil, halibut oil, menhaden oil, orange roughy oil and shark liver oil.

77. (Previously Presented) The method of claim 37, wherein the immunological adjuvant comprises an oil component.

78. (Previously Presented) The method of claim 77, wherein the oil component is selected from the group consisting of a single oil, and a mixture of oils.

79-82. (Cancelled).

83. (Previously Presented) The method of claim 37, wherein the immunological adjuvant comprises Mycobacterial cell wall extract.

84. (Previously Presented) The method of claim 37, wherein the immunological adjuvant comprises Mycobacterial DNA.

85. (Previously Presented) The method of claim 37, wherein the immunological adjuvant comprises a Mycobacterial cell wall complex.

86. (Previously Presented) The method of claim 63, wherein the aliphatic nitrogenous base is selected from the group consisting of an amine, a quaternary ammonium compound, a guanidine, a benzamidine, and a thiouronium.

87. (Previously Presented) The method of claim 37, wherein the immunological adjuvant comprisesdimethyl-dioctadecylammonium bromide.

88. (Previously Presented) The method of claim 63, wherein the aliphatic nitrogenous base is N,N-dioctadecyl-N,N-bis(2-hydroxyethyl)propanediamine.

89. (Previously Presented) The method of claim 46, wherein EspA+Tir comprise 20% of EHEC protein in the cell culture supernatant present in the composition.

90. (Previously Presented) The method of claim 35, wherein the ruminant is an ovine subject.

91. (Previously Presented) The method of claim 51, wherein the immunological adjuvant comprises an agent selected from the group consisting of an emulsifying agent, a muramyl dipeptide, an aqueous agent, a chitosan-based agent, a saponin, an oil, a lipopolysaccharide, a bacterial cell wall extract, a bacterial DNA, a bacterial complex, a synthetic oligonucleotide, and a aliphatic nitrogenous base.

92. (Previously Presented) The method of claim 51, wherein the immunological adjuvant comprises an oil component.

93. (Previously Presented) The method of claim 56, wherein the non-oil-in-water emulsion is selected from the group consisting of an oil emulsion, a water-in-oil emulsion, and a water-in-oil-in-water emulsion.

94. (Previously Presented) The method of claim 51, wherein the immunological adjuvant comprises Mycobacterial cell wall extract.

95. (Previously Presented) The method of claim 51, wherein the immunological adjuvant comprises Mycobacterial DNA.

96. (Previously Presented) The method of claim 51, wherein the immunological adjuvant comprises a Mycobacterial cell wall complex.

97. (Previously Presented) The method of claim 51, wherein the immunological adjuvant comprises Mycobacterial cell wall extract.

98. (Previously Presented) The method of claim 51, wherein the immunological adjuvant comprises Mycobacterial DNA.

99. (Previously Presented) The method of claim 51, wherein the immunological adjuvant comprises a Mycobacterial cell wall complex.

100. (Previously Presented) The method of claim 51, wherein the immunological adjuvant comprises dimethyl-dioctadecylammonium bromide.

101. (Previously Presented) The method of claim 48, wherein EspA+Tir comprise 20% of EHEC protein in the cell culture supernatant present in the composition.

102. (Previously Presented) The method of claim 60, wherein EspA+Tir comprise 20% of EHEC protein in the cell culture supernatant present in the composition.

103. (Previously Presented) The method of claim 62, wherein EspA+Tir comprise 20% of EHEC protein in the cell culture supernatant present in the composition.

104. (Previously Presented) The method of claim 49, wherein the ruminant is an ovine subject.

105. (Previously Presented) The method of claim 33, wherein the cell culture supernatant is concentrated.

106. (Previously Presented) The method of claim 34, wherein the cell culture supernatant is concentrated.

107. (Previously Presented) A method for reducing colonization of enterohemorrhagic *Escherichia coli* (EHEC) in a non-human mammal comprising administering to said non-human mammal an effective amount of a composition comprising an isolated EHEC cell culture supernatant containing one or more EHEC serotypes wherein the cell culture supernatant is produced by the process of incubating the cell culture in media comprising minimal media supplemented with 0.1% Casamino Acids, 0.4% glucose, 8 mM $MgSO_4$ and 44 mM $NaHCO_3$.

108. (Previously Presented) The method of claim 107, wherein the cell culture supernatant is concentrated.

109. (Previously Presented) The method of claim 107, wherein the one of more EHEC serotypes are selected from the group consisting of O157, O158, O5, O8, O18, O26, O45, O48, O52, O55, O75, O76, O78, O84, O91, O103, O104, O111, O113, O114, O116, O118, O119, O121, O125, O28, O145, O146, O163, and O165.

110. (Previously Presented) The method of claim 107, wherein the step of administering is by a route selected from the group consisting of oral, subcutaneous, intramuscular, intravenous, subcutaneous, intradermal, transdermal and subdermal.

111. (Previously Presented) A method for reducing shedding of enterohemorrhagic *Escherichia coli* (EHEC) from a non-human mammal comprising administering to said non-human mammal an effective amount of a composition comprising an isolated EHEC cell culture supernatant containing one or more EHEC serotypes wherein the cell culture supernatant is produced by the process of incubating the cell culture in media comprising minimal media supplemented with 0.1% Casamino Acids, 0.4% glucose, 8 mM $MgSO_4$ and 44 mM $NaHCO_3$.

112. (Previously Presented) The method of claim 111, wherein the cell culture supernatant is concentrated.

113. (Previously Presented) The method of claim 111, wherein the one of more EHEC serotypes are selected from the group consisting of O157, O158, O5, O8, O18, O26, O45, O48, O52, O55, O75, O76, O78, O84, O91, O103, O104, O111, O113, O114, O116, O118, O119, O121, O125, O28, O145, O146, O163, and O165.

114. (Previously Presented) The method of claim 111, wherein the step of administering is by a route selected from the group consisting of oral, subcutaneous, intramuscular, intravenous, subcutaneous, intradermal, transdermal and subdermal.

115. (Previously Presented) The method of claim 33, wherein the step of administering is by a route selected from the group consisting of oral, subcutaneous, intramuscular, intravenous, subcutaneous, intradermal, transdermal and subdermal.

116. (Previously Presented) The method of claim 34, wherein the step of administering is by a route selected from the group consisting of oral, subcutaneous, intramuscular, intravenous, subcutaneous, intradermal, transdermal and subdermal.